



## Complete Genome Sequence of Campylobacter fetus subsp. testudinum Strain Pet-3, Isolated from a Lizard (Hydrosaurus pustulatus)

© Chao-Min Wang,<sup>a</sup> Zong-Yen Wu,<sup>b</sup> Wei-Yau Shia,<sup>b</sup> Yi-Jyun Jhou,<sup>c</sup> Kwong-Chung Tung,<sup>b</sup> Ching-Lin Shyu<sup>b</sup>

Research Center for Biodiversity, China Medical University, Taichung, Taiwan<sup>a</sup>; Department of Veterinary Medicine, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute of Microbiology and Public Health, College of Veterinary Medicine, National Chung-Hsing University, Taichung, Taiwan<sup>b</sup>; Graduate Institute Organ

The whole-genome sequence for *Campylobacter fetus* subsp. *testudinum*, a pathogen isolated from humans and turtles, has been reported recently. We present another completed genome sequence of the *C. fetus* subsp. *testudinum* strain pet-3, which was isolated from a lizard in Taiwan, for further genomic comparison study.

Received 19 December 2014 Accepted 8 January 2015 Published 19 February 2015

Citation Wang C-M, Wu Z-Y, Shia W-Y, Jhou Y-J, Tung K-C, Shyu C-L. 2015. Complete genome sequence of Campylobacter fetus subsp. testudinum strain Pet-3, isolated from a lizard (Hydrosaurus pustulatus). Genome Announc 3(1):e01420-14. doi:10.1128/genomeA.01420-14.

Copyright © 2015 Wang et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license.

Address correspondence to Chao-Min Wang, leo8wang@yahoo.com.tw, or Ching-Lin Shyu, clshyu@nchu.edu.tw.

"ampylobacter fetus is an important human pathogen with a diverse host range, including mammals, fowls, and reptiles (1-3). Previous studies demonstrated that strains of C. fetus isolated from reptiles were genetically distinct from mammalassociated C. fetus by multilocus sequencing typing (4, 5). The genetic distance between mammal- and reptile-associated C. fetus is larger than within mammal-associated C. fetus. Recently, a polyphasic study was undertaken to determine the taxonomic position of those strains isolated from reptiles and humans (6). The results, including a whole-genome sequence, showed that those strains are closely related to C. fetus but are clearly different from recognized subspecies of *C. fetus*. Therefore, a novel subspecies, *C. fetus* subsp. testudinum, is proposed (7). Here, we report a whole-genome sequence of C. fetus subsp. testudinum strain Pet-3, which was isolated from a lizard (Hydrosaurus pustulatus), for further genomic comparison study.

Whole-genome paired-end sequencing was performed on an Illumina MiSeq desktop sequencer (Illumina Co., USA). A total of 14,590,400 reads were generated, resulting in 2,045-fold sequencing coverage. After end-trimming by Q20 cutoff, the remaining reads were *de novo* assembled into 35 contigs using CLC Genomics Workbench version 6.5.1. By comparing the genome sequences from related species using MUMmer version 3.23 and further bioinformatics analysis using GapCloser version 1.12-r6, these 35 contigs were manually inspected and concatenated into a single scaffold of a draft genome sequence using Consed version 26. The remaining six gaps within the scaffold were further closed by PCR walking and Sanger sequencing.

The whole-genome size of *C. fetus* subsp. *testudinum* Pet-3 is 1,776,391 bp, with an average G+C content of 33.13%. The open reading frames (ORFs) were predicted using Glimmer version 3.02 and prokaryotic GeneMark.hmm version 2.10f. The rRNA (rRNA) and tRNA (tRNA) genes were identified by RNAmmer version 1.2 and tRNAscan-SE version 1.23 software, respectively. The genome contains 1,796 putative protein-coding genes, 3 rRNA operons, and 43 tRNA genes. The putative functions of the genes were annotated against the NCBI nr (nonredundant), mi-

crobial RefSeq protein, and COG (Clusters of Orthologous Groups for Unicellular clusters) databases, as well as the KEGG (Kyoto Encyclopedia of Genes and Genomes) protein database using BLASTp. BLAST analysis indicated a high degree of similarity between the reptile-associated C. fetus subsp. testudinum genomes. A clustered regularly interspaced short palindromic repeats (CRISPR)-Cas system, an S-layer coding region, and a putative tricarballylate catabolism pathway were presented as predicted by protein analyses. Additionally, based on the core proteomes, 99% to 100% amino acid identity was observed between the proteomes common to C. fetus subsp. testudinum strain 03-427 and strain Pet-3. The most variable region between two strains was observed in the coding region of surface array protein A, with only 83% to 91% similarity. Further study on comparing the whole-genome sequence between C. fetus subsp. testudinum strains can provide a better understanding of the host virulence, adaptation evolution, and the taxonomic structure for those reptile-associated Campylobacter subspecies.

**Nucleotide sequence accession number.** The complete genome sequence of *C. fetus* subsp. *testudinum* strain Pet-3 has been deposited in GenBank under the accession number CP009226.

## **ACKNOWLEDGMENTS**

This work was supported in part by the Council of Agriculture Taiwan (102 AS-1.1.2-BQ-B3), the Ministry of Science and Technology (MOST 103-2811-B-039-020), and the Ministry of Education Taiwan under the Aim for the Top University plan.

## REFERENCES

- 1. **Torphy DE, Bond WW**. 1979. *Campylobacter fetus* infections in children. Pediatrics **64**:898–903.
- Sauerwein RW, Bisseling J, Horrevorts AM. 1993. Septic abortion associated with Campylobacter fetus subspecies fetus infection: case report and review of the literature. Infect 21:331–333. http://dx.doi.org/10.1007/BF01712458.
- 3. Tu ZC, Zeitlin G, Gagner JP, Keo T, Hanna BA, Blaser MJ. 2004. *Campylobacter fetus* of reptile origin as a human pathogen. J Clin Microbiol 42:4405–4407. http://dx.doi.org/10.1128/JCM.42.9.4405-4407.2004.

- Wang CM, Shia WY, Jhou YJ, Shyu CL. 2013. Occurrence and molecular characterization of reptilian *Campylobacter fetus* strains isolated in Taiwan. Vet Microbiol 164:67–76. http://dx.doi.org/10.1016/j.vetmic.2013.01.008.
- Dingle KE, Blaser MJ, Tu ZC, Pruckler J, Fitzgerald C, van Bergen MA, Lawson AJ, Owen RJ, Wagenaar JA. 2010. Genetic relationships among reptilian and mammalian *Campylobacter fetus* strains determined by multilocus sequence typing. J Clin Microbiol 48:977–980. http://dx.doi.org/ 10.1128/JCM.01439-09.
- 6. Fitzgerald C, Tu ZC, Patrick M, Stiles T, Lawson AJ, Santovenia M,
- Gilbert MJ, van Bergen M, Joyce K, Pruckler J, Stroika S, Duim B, Miller WG, Loparev V, Sinnige JC, Fields PI, Tauxe RV, Blaser MJ, Wagenaar JA. 2014. *Campylobacter fetus* subsp. *testudinum* subsp. nov., isolated from humans and reptiles. Int J Syst Evol Microbiol 64:2944–2948. http://dx.doi.org/10.1099/ijs.0.057778-0.
- 7. Gilbert MJ, Miller WG, Yee E, Blaser MJ, Wagenaar JA, Duim B. 2013. Complete genome sequence of *Campylobacter fetus* subsp. *testudinum* strain 03–427<sup>T</sup>. Genome Announc 1(6):e01002-13. http://dx.doi.org/10.1128/genomeA.01002-13.